

What we claim is:

1. An apparatus for processing compounds in small volumes by electrophoretic separation, the apparatus comprising:

- 5 (a) a cathode in a static cathode buffer zone;
- (b) an anode in a static anode buffer zone, the anode disposed relative to the cathode so as to be adapted to generate an electric field in an electric field area therebetween upon application of a voltage potential between the cathode and anode;
- (c) a first separation barrier disposed in the electric field area;
- 10 (d) a second separation barrier disposed between a selected one of the cathode buffer zone and the anode buffer zone and the first barrier so as to define a first interstitial volume therebetween;

wherein in use, electrophoretic buffer is disposed in the cathode buffer zone and the anode buffer zone, a sample constituent is provided to the first interstitial volume;

15 wherein upon application of the voltage potential, a selected separation product is removed from the sample constituent through a selected one of the first and second separation barriers, and provided to a selected one of the cathode buffer and anode buffer zones; and wherein there is substantially no circulation of buffer or sample constituent in the buffer zones or the first interstitial volume.

20

2. The apparatus according to claim 1 wherein the small volume is less than about 5 mL.

3. The apparatus according to claim 2 wherein the small volume is up to about 2 mL.

25

4. The apparatus according to claim 3 wherein the small volume is from about 0.02 mL to about 0.1 mL.

30 5. The apparatus according to claim 1 wherein a ratio of sample to barrier surface area is less than about 1 mL/cm².

6. The apparatus according to claim 5 wherein the ratio is up to about 0.5 mL/cm².

7. The apparatus according to claim 6 wherein the ratio is up to about 0.1 mL/cm².

8. The apparatus according to claim 7 wherein the ratio is about 0.02 mL/cm².

9. The apparatus according claim 1 wherein the first and second barriers are membranes and wherein such membranes are selected from the group consisting of electrophoresis separation membranes having a defined pore size or restriction membranes which allow flow of ions into and out of a chamber or compartment under the influence of an electric field but do not allow movement of macromolecules, or a combination thereof.

10. The apparatus according to claim 9 wherein at least one of the first and second membranes is an ion-permeable electrophoresis separation membrane comprised of polyacrylamide and having a molecule mass cut-off from about 1 kDa to about 1500 kDa.

11. The apparatus according to claim 10 wherein a selected one of the first and second membranes is an ion-permeable electrophoresis separation membrane comprised of polyacrylamide and having a molecule mass cut-off from about 1 kDa to about 1500 kDa and the other membrane is a restriction membrane comprised of polyacrylamide and having a molecular mass cut-off less than the separation membrane.

12. The apparatus according claim 1 wherein the first and second separation barriers are membranes and such membranes are comprised of materials selected from the group consisting of ultrafiltration materials, electrodialysis materials, haemodialysis materials, and combinations thereof.

13. The apparatus according to claim 1 wherein the first and second separation barriers are adapted to be removed from the apparatus.

14. The apparatus according to claim 1 comprising a plurality of separation
5 barriers spaced apart defining a plurality of interstitial volumes.

15. An apparatus for processing compounds in small volumes by electrophoretic separation, the apparatus comprising:

- (a) a cathode in a static cathode buffer zone;
- 10 (b) an anode in a static anode buffer zone, the anode disposed relative to the cathode so as to be adapted to generate an electric field in an electric field area therebetween upon application of a voltage potential between the cathode and anode;
- (c) a first separation barrier disposed in the electric field area;
- (d) a second separation barrier disposed between the cathode buffer zone and
15 the first barrier so as to define a first interstitial volume therebetween;
- (e) a third separation barrier disposed between the anode buffer zone and the first barrier so as to define a second interstitial volume therebetween;

wherein in use, electrophoretic buffer is disposed in the cathode buffer zone, the anode buffer zone and at least one of the first interstitial and second interstitial volumes, a
20 sample constituent is provided to a selected one of the first interstitial and second interstitial volumes; wherein upon application of the voltage potential, a selected separation product is removed from the sample constituent, through the first separation barrier, and provided to the other of the first interstitial and second interstitial volumes; and wherein there is substantially no circulation of buffer or sample constituent in the
25 buffer zones, the first interstitial volume or the second interstitial volume.

16. The apparatus according to claim 15 wherein the small volume is less than about 5 mL.

17. The apparatus according to claim 15 wherein the small volume is up to
30 about 2 mL.

18. The apparatus according to claim 17 wherein the small volume is from about 0.02 mL to about 0.1 mL.

19. The apparatus according claim 15 wherein a ratio of sample to barrier
5 surface area is less than about 1 mL/cm².

20. The apparatus according to claim 19 wherein the ratio is up to about 0.5 mL/cm².

21. The apparatus according to claim 20 wherein the ratio is up to about 0.1 mL/cm².

22. The apparatus according to claim 21 wherein the ratio is about 0.02 mL/cm².

23. The apparatus according to claim 15 wherein the first separation barrier is an electrophoresis membrane having a defined pore size and the second and third separation barriers are restriction membranes which allow flow of ions into and out of a chamber or compartment under the influence of an electric field.

24. The apparatus according to claim 23 wherein the separation membrane is an ion-permeable electrophoresis separation membranes comprised of polyacrylamide and having a molecule mass cut-off from about 1 kDa to about 1500 kDa.

25. The apparatus according to claim 24 wherein the restriction membrane is comprised of polyacrylamide and having a molecular mass cut-off less than the separation membrane.

26. The apparatus according claim 15 wherein the first, second and third
30 separation barriers are membranes and such membranes comprised of materials selected from the group consisting of ultrafiltration materials, electro dialysis materials, haemodialysis materials, and combinations thereof.

27. The apparatus according to claim 15 wherein the first, second and third separation barriers membranes are adapted to be removed from the apparatus.

5 28. An apparatus for processing compounds in small volumes by electrophoretic separation, the apparatus comprising:

- (a) a cathode in a static cathode buffer zone;
- (b) an anode in a static anode buffer zone, the anode disposed relative to the cathode so as to be adapted to generate an electric field in an electric field area
- 10 therebetween upon application of a voltage potential between the cathode and anode;
- (c) a first separation barrier disposed in the electric field area;
- (d) a second separation barrier disposed between the cathode buffer zone and the first barrier so as to define a first sample interstitial volume therebetween;
- (e) a third separation barrier disposed between the anode buffer zone and the
- 15 first barrier so as to define a first separation interstitial volume therebetween;

wherein in use, electrophoretic buffer is disposed in the cathode buffer zone, the anode buffer zone and at least one of the first sample interstitial and first separation interstitial volumes, a sample constituent is provided to the first sample interstitial volume; wherein upon application of the voltage potential, a selected separation product is

20 removed from the sample constituent, through the first separation barrier, and provided to the first separation interstitial volume; and wherein there is substantially no circulation of buffer or sample constituent in the buffer zones, the first sample interstitial volume or the first separation interstitial volume.

25 29. The apparatus according to claim 28 wherein a least fourth separation barrier is disposed between the second separation barrier and the cathode buffer zone to define at least a second sample interstitial volume wherein sample constituents are provided to selected sample interstitial volumes, wherein upon application of the voltage potential, a selected separation product is removed from the sample constituents, through

30 the separation barriers, and provided to the first separation interstitial volume.

30. The apparatus according to claim 29 wherein at least a fifth separation barrier is disposed between the third separation barrier and the anode buffer zone to define at least a second separation interstitial volume, wherein sample constituents are provided to selected sample interstitial volumes, wherein upon application of the voltage potential, a selected separation product is removed from the sample constituents, through the separation barriers, and provided to selected separation interstitial volumes.

31. The apparatus according to claim 30 wherein the apparatus further comprises at least a second cathode in a second static cathode buffer zone and a second anode in second static anode zone disposed relative to the second cathode so as to be adapted to generate an electric field between selected sample interstitial volumes and selected separation interstitial volumes.

32. The apparatus according to claim 28 wherein at least a fifth separation barrier is disposed between the third separation barrier and the anode buffer zone to define at least a second separation interstitial volume, wherein a sample constituent is provided to the first sample interstitial volumes, wherein upon application of the voltage potential, a selected separation product is removed from the sample constituent, through the separation barriers, and provided to selected separation interstitial volumes.

33. A method for de-salting or dialysing a small volume sample containing at least one compound, the method comprising:

- (a) providing an apparatus according to claim 1;
- (b) adding buffer to the cathode and anode buffer zones;
- (c) placing a sample in the first interstitial volume; and
- (d) applying a voltage potential, wherein upon application of the voltage potential, selected salts in the sample are removed from the mixture through a selected one of the first and second separation barriers and provided to a selected one of the cathode buffer zone and the anode buffer zone, wherein at least one compound is substantially retained in the first interstitial volume, and wherein there is substantially no circulation of buffer or sample constituent in the buffer zones and the first interstitial volume.

34. A method for de-salting or dialysing a small volume sample containing at least one compound, the method comprising:

- (a) providing an apparatus according to claim 15;
- 5 (b) adding buffer to the cathode and anode buffer zones and to at least one of the interstitial volumes;
- (c) placing a sample in a selected one of the first interstitial and second interstitial volumes; and
- (d) applying a voltage potential between the first interstitial and second
- 10 interstitial volumes, wherein upon application of the voltage potential, selected salts in the sample are removed from the mixture through a selected one of the first separation barrier and provided to the other of the first interstitial and second interstitial volume, wherein at least one compound is substantially retained in first interstitial volume from which the selected salts were removed, wherein there is substantially no circulation of buffer or
- 15 sample constituent in the buffer zones, the first interstitial volume, and the second interstitial volume.

35. A method of separating a compound in small volumes of solution by electrophoretic separation, the method comprising:

- 20 (a) providing an apparatus according to claim 1;
- (b) adding buffer to the cathode and anode buffer zones;
- (c) placing a sample in the first interstitial volume; and
- (d) applying a voltage potential, wherein upon application of the voltage potential, selected separation products in the sample are removed from the mixture
- 25 through a selected one of the first and second separation barriers and provided to a selected one of the cathode buffer zone and the anode buffer zone, wherein at least one compound is substantially retained in the first interstitial volume, and wherein there is substantially no circulation of buffer or sample constituent in the buffer zones and the first interstitial volume.

36. A method of separating a compound in small volumes of solution by electrophoretic separation, the method comprising:

(a) providing an apparatus according to claim 15;
(b) adding buffer to the cathode and anode buffer zones and to at least one of the interstitial volumes;

(c) placing a sample in a selected one of the first interstitial and second
5 interstitial volumes; and

(d) applying a voltage potential between the first interstitial and second interstitial volumes, wherein upon application of the voltage potential, selected separation products in the sample are removed from the mixture through a selected one of the first separation barrier and provided to the other of the first interstitial and second interstitial
10 volume, wherein at least one compound is substantially retained in first interstitial volume from which the selected separation products were removed, wherein there is substantially no circulation of buffer or sample constituent in the buffer zones, the first interstitial volume, and the second interstitial volume.

05837678 "041801